

CLAIMS

1. An isolated, natural or synthetic polynucleotide characterized in that it comprises at least the 5 three open reading frames corresponding to the sequences SEQ ID No.1, SEQ ID No.2 and SEQ ID No.3.

2. The polynucleotide as claimed in claim 1, characterized in that it also comprises the open reading frame corresponding to the sequence SEQ ID 10 No.4.

3. An isolated, natural or synthetic polynucleotide characterized in that it corresponds to the sequence SEQ ID No.5.

4. An isolated, natural or synthetic polynucleotide 15 characterized in that it comprises at least one of the three open reading frames corresponding to the sequences SEQ ID No.2, SEQ ID No.3 and SEQ ID No.4.

5. An isolated, natural or synthetic polynucleotide corresponding to any one of the sequences SEQ ID 20 No.2, SEQ ID No.3 or SEQ ID No.4.

6. A vector, characterized in that it comprises one of the polynucleotides as claimed in any one of claims 1 to 5.

7. The vector as claimed in claim 6, characterized 25 in that it is a plasmid, a cosmid, a bacterial artificial chromosome (BAC), an integrative element of actinobacteria, a virus or else a bacteriophage.

8. The use of at least one of the polynucleotides 30 as claimed in any one of claims 1 to 5 or of one of its fragments of at least 15 nucleotides or of one of the vectors as claimed in either one of claims 6 or 7, as a probe.

9. The use of at least one of the polynucleotides 35 as claimed in any one of claims 1 to 5 or of one of its fragments of at least 15 nucleotides or of one of the vectors as claimed in either one of claims 6 or 7, as a primer for amplifying nucleic acid sequences.

10. An isolated, natural or synthetic poly-

polypeptide characterized in that it comprises at least any one of the sequences SEQ ID No.7 to SEQ ID No.10.

11. An isolated, natural or synthetic polypeptide characterized in that it corresponds to any one of the sequences SEQ ID No.7 to SEQ ID No.10.

12. An isolated, natural or synthetic polypeptide characterized in that it is encoded by one of the polynucleotides as claimed in any one of claims 1 to 5 or one of the vectors as claimed in either one of claims 6 or 7.

13. The use of a polynucleotide as claimed in any one of claims 1 to 5 or of a vector as described in either one of claims 6 or 7, for preparing a polypeptide as described in any one of claims 10 to 12.

14. The use, particularly *in vitro*, of at least one polypeptide as claimed in any one of claims 10 to 12, alone or in combination, for preparing cyclo-dipeptides and/or diketopiperazine derivatives substituted in the 3- and 6-positions with α,β -unsaturated amino acid side chains, particularly albonoursin.

15. The use of at least one polynucleotide as claimed in any one of claims 1 to 5 or of a vector as described in either one of claims 6 or 7, for preparing a modified biological system or a modified *in vitro* acellular system.

16. The use as claimed in claim 15, characterized in that the modified biological system is a microorganism or a heterologous expression system using prokaryotes or eukaryotes as hosts.

17. A modified biological system, characterized in that it contains at least one of the polynucleotides as described in any one of claims 1 to 5 and/or at least one of the vectors as described in either one of claims 6 or 7.

18. The biological system as claimed in claim 17, characterized in that it consists of a microorganism or a heterologous expression system using prokaryotes or eukaryotes as hosts, or else an *in vitro* acellular system.

19. The biological system as claimed in claim 18, characterized in that the microorganism is a bacterium such as *Escherichia coli* or *Streptomyces lividans*.

5 20. A modified *in vitro* acellular system, characterized in that it contains at least one of the polynucleotides as described in any one of claims 1 to 5 and/or at least one of the vectors as described in either one of claims 6 or 7.

10 21. The use of at least one modified biological system as claimed in any one of claims 17 to 19 or of a modified *in vitro* acellular system as claimed in claim 20, for preparing cyclodipeptides and/or diketopiperazine derivatives substituted in the 3- and 6-positions with α,β -unsaturated amino acid side chains, particularly albonoursin.

22. A method for synthesizing, *in vitro*, cyclodipeptides, characterized in that:

20 (1) two amino acids, which may be identical or different, are brought into contact, under suitable conditions, with the polypeptide AlbC (SEQ ID No.9), and

(2) the cyclodipeptide obtained is purified.

25 23. A method for synthesizing, *in vitro*, an α,β -unsaturated diketopiperazine derivative substituted in the 3- and 6-positions with amino acid side chains, characterized in that:

30 (1) two amino acids, which may be identical or different, are brought into contact, under suitable conditions, with the polypeptide AlbC (SEQ ID No.9) and the cyclodipeptide obtained is purified, and

(2) the cyclodipeptide obtained in step (1) is brought into contact with AlbA (SEQ ID No.6), AlbB1 (SEQ ID No.7) and AlbB2 (SEQ ID No.8) and the α,β -unsaturated diketopiperazine derivative obtained is purified.

24. The method as claimed in claim 23, characterized in that the method also comprises, in step (2), the polypeptide AlbD (SEQ ID No.10).

25. The method as claimed in any one of claims 22 to 24, characterized in that the amount of polypeptides is between 0.1 nM and 10 μ M, preferably between 10 nM and 1 μ M.

5 26. A method for synthesizing a cyclodipeptide, characterized in that:

10 (1) a biological system comprising at least the polynucleotide albC (SEQ ID No.3) is brought into contact under conditions suitable for culturing said chosen biological system, and

(2) the cyclodipeptide obtained is purified.

27. The method as claimed in claim 26, characterized in that the biological system also comprises the polynucleotide albD (SEQ ID No.4).

15 28. A method for synthesizing a diketopiperazine derivative substituted in the 3- and 6-positions with α,β -unsaturated amino acid side chains, characterized in that:

20 (1) a biological system comprising a polynucleotide comprising at least albA, albB and albC (SEQ ID Nos.1 to 3) is brought into contact under conditions suitable for culturing said chosen biological system, and

25 (2) the α,β -unsaturated diketopiperazine derivative obtained is purified.

29. The method as claimed in claim 28, characterized in that the biological system also comprises the polynucleotide albD (SEQ ID No.4).

30 30. The method as claimed in any one of claims 26 to 29, characterized in that the biological system is a microorganism, for instance a bacterium such as *Escherichia coli* or *Streptomyces lividans*, or any known heterologous expression system using prokaryotes or eukaryotes as hosts, or even an *in vitro* acellular system.

35 31. The method as claimed in any one of claims 22 to 30, characterized in that the amount of amino acids is between 0.1 mM and 100 mM, preferably between 1 mM and 10 mM.